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Maternal Levels of Perfluoroalkyl Substances (PFAS) during Early Pregnancy in Relation to Preeclampsia Subtypes and Biomarkers of Preeclampsia Risk

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Background: Prenatal exposure to perfluoroalkyl substances (PFAS) has been previously associated with preeclampsia, although findings are mixed with respect to the direction and magnitude of effect. To our knowledge, no studies have examined associations between PFAS and preeclampsia subtypes, which may have distinct etiologies.

Objective: We examined associations between PFAS, any preeclampsia diagnosis, and early- and late-onset preeclampsia. In addition, we estimated associations between PFAS and the angiogenic biomarkers soluble fms-like tyrosine kinase-1 (sFLT-1) and placental growth factor (PlGF), which provide an estimate of pro- and anti-angiogenic activity within the placenta.

Methods: This case–control study ($n = 75$ cases, $n = 75$ controls) was sampled from the LIFECODES birth cohort. Nine legacy PFAS were quantified in maternal plasma from early pregnancy (median = 10 wk) and angiogenic biomarkers were quantified in maternal plasma from four study visits (median = 10, 18, 26, and 35 wk). Logistic regression was used to estimate the odds ratios (ORs) and 95% confidence intervals (CIs) of the association between an interquartile range (IQR)-increase in PFAS and preeclampsia outcomes. Linear regression was used to estimate associations between an IQR-increase in PFAS and concentrations of angiogenic biomarkers.

Results: Both perfluorodecanoic acid (OR = 1.64, 95% CI: 1.08, 2.47) and perfluorooctanesulfonic acid (OR = 1.60, 95% CI: 1.06, 2.43) were associated with higher odds of late-onset preeclampsia. Associations tended to be below the null for early-onset preeclampsia, although findings were imprecise. Few associations were noted between PFAS and angiogenic biomarkers.

Discussion: Maternal PFAS concentrations were associated with higher odds of late-onset preeclampsia. Heterogeneity of preeclampsia should be considered in future studies because populations may have different distributions of disease subtypes. https://doi.org/10.1289/EHP9091

Introduction

Perfluoroalkyl substances (PFAS) are a set of synthetic chemicals that are used widely in commercial products and manufacturing (ITRC 2020a). These compounds are characterized by a perfluoroalkyl chain and a functional group (e.g., carboxylic, sulfonic acid) (ITRC 2020b). Because the half-lives of some of these chemicals are long, ranging from several days to over a decade (ATSDR 2021), and they do not degrade in the environment (Sunderland et al. 2019; Zhang et al. 2013), they are sometimes referred to as forever chemicals. As a result, PFAS are ubiquitous in the environment and are commonly detected in humans, both in the United States (Calafat et al. 2019) and globally (Göckener et al. 2020; Seo et al. 2018; Timmermann et al. 2020). Although some long-chain PFAS are being phased out of production (and also known as legacy PFAS), the use of other novel PFAS is increasing (ITRC 2020a). Exposure to PFAS has been linked to a wide array of adverse health outcomes, including reproductive outcomes (Blake and Fenton 2020). For example, prenatal PFAS exposure has been linked to excessive gestational weight gain (Ashley-Martin et al. 2016; Mitro et al. 2020), preterm birth (Meng et al. 2018; Waterfield et al. 2020), and preeclampsia (Savitz et al. 2012; Stein et al. 2009).

Preeclampsia is a reproductive disorder that affects ~2–8% of all pregnancies within the United States (Ananth et al. 2013) and represents a major contributor to maternal mortality (Steegers et al. 2010). Preeclampsia is defined as the presence of new or worsening hypertension and proteinuria after 20 wk of gestation (ACOG 2013). Notably, the underlying causes of preeclampsia remain poorly understood. This is in part because preeclampsia is a heterogenous syndrome, with diverse clinical and pathophysiologic presentations. For example, preeclampsia is classically characterized by the presence of an insufficient placenta, which is poorly perfused and hypoxic (Steegers et al. 2010). However, indicators of severe placental disease disproportionately impact preeclampsia cases that arise earlier during pregnancy (i.e., <34 wk gestation). Late-onset preeclampsia (i.e., ≥34 wk gestation) cases often have minimal placental involvement (Roberts and Hubel 2009; Steegers et al. 2010). Early-onset disease is typically considered more severe and is more strongly associated with adverse fetal and maternal outcomes than late-onset disease (Bellamy et al. 2007; Mongraw-Chaffin et al. 2010), although late-onset preeclampsia may still progress to severe disease (Kenneth et al. 2010). Given these observations, preeclampsia is thought to comprise different subtypes that may have distinct mechanistic underpinnings (Raymond and Peterson 2011; van der Merwe et al. 2010). Thus, it may be necessary to examine associations between environmental exposures and preeclampsia subtypes (i.e., early- and late-onset) separately (von Dadelszen et al. 2003).

Early results from the retrospective C8 Health Study, which includes a highly exposed population living in the Ohio River Valley, indicated that perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS) were associated with higher odds of self-reported preeclampsia (Savitz et al. 2012; Stein et al. 2009). However, more recent studies in populations with lower PFAS exposures have reported mixed associations with this outcome (Borghese et al. 2020; Huang et al. 2019; Huo et al. 2020; Rylander et al. 2020; Starling et al. 2014; Wikström et al. 2019).

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To our knowledge, previous studies have not examined PFAS exposure in relation to preeclampsia subtypes. Examining whether there are distinct associations between PFAS and preeclampsia subtypes may be important in understanding the heterogeneity in previous studies because the sampled populations may have different distributions of early- or late-onset preeclampsia cases. Furthermore, associations between PFAS exposure and circulating angiogenic biomarkers, such as soluble fms-like tyrosine kinase-1 (sFLT-1) or placental growth factor (PlGF), should be considered, given that toxicologic studies have demonstrated an influence of PFAS on angiogenic processes in the placenta (Pham et al. 2020; Poteser et al. 2020). These factors are released by the placenta into the maternal circulation, giving an indication of the balance of anti-angiogenic (i.e., sFLT-1) to pro-angiogenic (i.e., PlGF) activity within the placenta (Maynard et al. 2003; McElrath et al. 2012). Notably, these factors have been shown to predict preeclampsia onset (Steegers et al. 2010) and may prove to have clinical utility in assessing disease risk (ACOG 2020). Therefore, associations between PFAS and angiogenic biomarkers may shed light on both the risk of developing preeclampsia and the underlying mechanisms contributing to disease progression.

In the present study, we used a case–control population designed to examine associations between exposure to nine different legacy PFAS during early pregnancy and preeclampsia, including early- and late-onset preeclampsia subtypes. Given differences between early- and late-onset preeclampsia, we hypothesized that associations with PFAS exposure could vary according to disease subtype. In addition, we investigated whether PFAS were associated with levels of the circulating angiogenic biomarkers, sFLT-1 and PlGF, and the sFLT-1:PlGF ratio.

Methods

Study Design

In this study, we used a case–control population nested within the larger LIFECODES study, an ongoing prospective pregnancy cohort that began in 2006. The LIFECODES study is based at Brigham and Women's Hospital in Boston, Massachusetts, and has been described in depth elsewhere (McElrath et al. 2012). Briefly, women are eligible for the study if they are *a*) at least 18 years of age, *b*) sought prenatal care before 15 wk of gestation, and *c*) intended on delivering at Brigham and Women's Hospital. All participants provide informed consent. Detailed questionnaires about demographics and medical history are provided and maternal plasma samples are collected at the first study visit (median = 10 wk gestation). After enrollment, participants attend three more study visits (median = 18, 26, and 35 wk gestation), where plasma samples are also collected. All specimens are stored at −80°C until analysis. This study was approved by the institutional review board of Brigham and Women's Hospital.

This case–control study consisted of 150 participants ($n = 75$ preeclamptic cases; $n = 75$ non-preeclamptic controls). Participants were sampled from those recruited between 2006–2008 ($N = 1648$). First, cases were randomly sampled from those with preeclampsia. Next, controls were subsequently sampled from non-preeclamptic participants, with frequency matching based on gestational age at the time of the first study visit (±2 wk gestation). The total number of participants in this study was determined as a result of available funds for the cost of PFAS analysis rather than power calculations. Gestational age was estimated according to the American College of Obstetrics and Gynecologists (ACOG) using the last menstrual period with verification by ultrasound measures (ACOG 2017).

Definition of Preeclampsia

Preeclampsia was diagnosed according to ACOG guidelines at the time of recruitment: new or worsening hypertension (≥140 mmHg systolic or ≥90 mmHg diastolic blood pressure) and proteinuria (>300 mg/24 h or protein/creatinine ratio of >0.20) after 20 wk of gestation (ACOG 2013). All cases of preeclampsia were deidentified and reviewed by a panel of two Maternal–Fetal Medicine–certified physicians. If consensus on diagnosis could not be reached, a third Maternal–Fetal Medicine–certified physician reviewed the case, and they met as a committee to resolve the conflict (necessary in ∼12% of all cases in LIFECODES). Gestational age at disease onset was recorded. Cases were further categorized as early- ($n = 21$) or late-onset ($n = 54$), depending on whether disease onset occurred prior to 34 wk gestation (von Dadelszen et al. 2003).

Covariates

Information on potential covariates was collected using questionnaires administered at the first study visit (i.e., enrollment), including maternal sociodemographics (e.g., race and ethnicity, educational attainment, insurance status). The questionnaire asked about several racial identities (i.e., "Caucasian," "Black," "South Asian," "East Asian," "Native American/Pacific Islander," "More than one race," "Other," and "Unsure") and participants were permitted to select multiple responses and to provide free-form text responses. Hispanic ethnicity was assessed separately. Behavioral information (e.g., smoking, alcohol use) and health information (e.g., personal health history, parity) was also self-reported on questionnaires. Self-reported health information was validated by two Maternal–Fetal Medicine specialists reviewing electronic medical records. Routine clinical information (e.g., maternal weight or blood pressure) was also collected at study visits. Body mass index (BMI) was calculated using self-reported maternal prepregnancy weight and height measured at the first study visit. Potential confounders were identified using a directed acyclic graph (DAG; Figure S1). Based on DAG analysis, the following variables were determined to be a sufficient adjustment set: maternal age, maternal BMI, maternal race and ethnicity, maternal education, maternal insurance status, and parity. We additionally considered whether smoking served as a potential confounder. Factors were retained in the final models if their exclusion influenced estimates by >10% relative to the sufficient adjustment set.

Quantification of Angiogenic Biomarkers

As previously described, levels of circulating maternal sFLT-1 and PlGF were measured in maternal plasma samples collected at each of the study visits (McElrath et al. 2012). Both sFLT-1 and PlGF were measured using ARCHITECT immunoassays (Abbott Laboratory). Unbound PlGF concentrations from 1 to 1,500 pg/mL were measured and total sFLT-1 concentrations from 0.10 to 150 ng/mL were measured. One sample had levels of sFLT-1 below the limit of detection (LOD) and was imputed as the LOD divided by the square root of 2 (Hornung and Reed 1990). The ratio of sFLT-1 to PlGF was also calculated.

Quantification of Perfluoroalkyl Substances

Maternal plasma concentrations of nine legacy PFAS [perfluoroheptanoic acid (PFHpA), PFOA, perfluorononanoic acid (PFNA), perfluorodecanoic acid (PFDA), perfluoroundecanoic acid (PFUnDA), perfluorohexane sulfonic acid (PFHxS), PFOS, 2-(*N*-methyl-perfluorooctane sulfonamido) acetic acid (MeFOSAA), and perfluorooctane sulfonamide (PFOSA)] were measured in samples collected at the first study visit

(median = 10 wk gestation) by NSF International (Ann Arbor, MI). Concentrations of PFAS were measured using a method developed to replicate the Centers for Disease Control and Prevention (CDC) polyfluoroalkyl chemicals Method Laboratory Procedure 6304.1 (Kuklenyik et al. 2005). Briefly, samples were acidified prior to concentration, separation, and detection via online solid-phase extraction (SPE) coupled to high-performance liquid chromatography (HPLC)-isotope dilution tandem mass spectrometry (LC-MS-MS). Specifically, samples were concentrated by on-line SPE interfaced with a Thermo Scientific Transcend system using a Cyclone-P extraction column. The analytes were then separated and focused using a Dionex UltiMate 3000 ultra-HPLC system with a reversed-phase chromatography using a Waters XBridge C18 analytical column. Last, chemicals were detected using a Thermo Scientific Transcend TXII Turbulent Flow system interfaced with a Thermo Scientific Quantiva MS. This method was validated and found to perform within acceptance criteria established within the CDC method. Levels below the LOD were replaced with the LOD divided by the square root of 2 (Hornung and Reed 1990). Eight of the measured PFAS were detected in at least 80% of the study participants and were used in further analyses.

Statistical Analysis

Statistical analyses were performed using SAS (version 9.4; SAS Institute Inc.). Demographic characteristics of participants were summarized in the overall cohort and according to case status. The trajectories of angiogenic biomarker concentrations across pregnancy were visualized using scatter plots with locally estimated scatter smoothing. The geometric mean (GM) and the 25th, 50th, 75th, and 95th percentiles of plasma PFAS concentrations and angiogenic biomarkers were also calculated for the overall cohort and according to case status and were contrasted using Wilcoxon rank sum or Kruskal-Wallis tests, where appropriate. Plasma PFAS concentrations measured in the LIFECODES cohort were compared with measures reported in reproductive-age women in the National Health and Nutrition Examination Survey (NHANES; cycle years: 2007–2008) (CDC 2019). Last, median PFAS concentrations were calculated according to demographic characteristics. These concentrations were contrasted across demographic characteristics using an analysis of variance following log-transformation of the PFAS concentrations because of their skewed distribution.

In our primary analyses, we used logistic regression models to estimate the odds ratio (OR) and 95% confidence interval (CI) of preeclampsia for an interquartile range (IQR)-increase in the concentrations of each PFAS. Logistic regression models were also used to separately estimate the ORs (95% CIs) of early- and late-onset preeclampsia. Given the matching design of this study, all models were adjusted for gestational age (in weeks) at the first study visit, when plasma samples were collected. In addition, the following covariates were included in our final models: maternal age (in years), prepregnancy BMI (in kilograms per meter squared), maternal race (non-Black/Black), parity (nulliparous/parous), maternal education (high school or less/some college or technical school/college or greater), and health insurance (private insurance/self-pay or public insurance). Because results were similar when adjusting for maternal race and ethnicity as a collapsed two-category variable (non-Black/Black), this collapsed variable was used instead of the four-category race and ethnicity variable (non-Hispanic White/Black/Hispanic/other) (Table S1). Given that missing data was minor (Table 1), a complete case analysis was performed in all subsequent analyses.

Although the case–control design of this study is most efficiently analyzed using logistic regression models, we conducted a secondary analysis of preeclampsia using Cox proportional hazards regression, which is better suited for analyzing preeclampsia outcomes given that it accounts for pregnancy duration and censoring due to preterm birth (Weinberg et al. 2017). Cox models were used to estimate the hazard ratios (HRs) and 95% CIs of preeclampsia. To account for the case–control sampling, we calculated and applied inverse probability weights (IPWs) based on the sampling fractions for cases and controls from the underlying LIFECODES cohort (case weight = 1.28, control weight = 17.5). Standard errors (SEs) were estimated using robust sandwich variance estimation. These models were constructed using the same covariates as the logistic regression models. The proportional hazards assumption was assessed and verified by creating time-dependent variables for each predictor in the model and testing for significance (Table S2).

Linear regression models were used to estimate the association between an IQR-increase in individual plasma PFAS concentrations and the concentrations of maternal angiogenic biomarkers at each study visit. Again, IPWs were applied to these models and robust sandwich variance estimation was used to estimate the SE and appropriately account for the case–control study design. Angiogenic biomarkers were ln-transformed prior to analysis because these measures were right skewed. The same covariates from our logistic regression models were used to ensure comparability between analyses. For models examining angiogenic biomarker concentrations at the first study visit, gestational age at the time of sample collection was represented using quadratic coding given the nonlinear association between gestational age and angiogenic biomarker concentrations at that time point (Figure 1). For all other models, gestational age was represented using a simple linear term. To increase the interpretability of the results, beta estimates and SEs from these regression models were converted to a percentage change value (95% CI) associated with an IQR-increase in individual PFAS concentrations using the following formula: percentage change = $(e^{\beta}-1)\times 100$.

Two sensitivity analyses were performed. Approximately 6% of the overall cohort had chronic hypertension. Chronic hypertension can complicate the diagnosis of preeclampsia, particularly when it co-occurs with kidney dysfunction (ACOG 2013). Therefore, as a sensitivity analysis, all regression models were reconstructed with additional adjustment for chronic hypertension (no/yes). In addition, given that previous studies on this topic have restricted their analyses to nulliparous women (Huo et al. 2020; Starling et al. 2014; Wikström et al. 2019), we replicated our analysis on nulliparous women in this cohort. Statistical significance throughout all analyses was defined as $p < 0.05$.

Results

Study Population

Demographic characteristics of the cohort are shown in Table 1 and were consistent with those in the underlying LIFECODES population (Honigberg et al. 2016). In general, participants who developed preeclampsia tended to have a higher prepregnancy BMI (29.3 vs. 24.3 kg/m^2) and were more likely to self-identify as Black (31% vs. 12%) and to be nulliparous (52% vs. 40%). They were also less likely to have a college education (57% vs. 74%) and to have access to private health insurance (74% vs. 86%). In addition, participants who developed preeclampsia more frequently reported a previous diagnosis of chronic hypertension (19% vs. 5%) and having had preeclampsia in a previous pregnancy (13% vs. 1%). We observed no meaningful difference in self-reported smoking during pregnancy between participants who did and did not develop preeclampsia (9% vs. 7%). Characteristics are further broken down according to early- and late-onset preeclampsia in Table S3. Participants who developed late-onset preeclampsia were less likely to have chronic

Table 1. Baseline demographics in the overall LIFECODES case–control study population and according to case status, recruited from 2006 to 2008.

Population characteristics	Median (25th–75th percentile) or *n* (%)		
	Total weighted population ($n=150$)[a]	Controls ($n=75$)	Preeclampsia cases ($n=75$)
Maternal age (y)	32.2 (29.2–36.2)	32.2 (29.4–36.3)	32.6 (27.9–36.2)
Maternal prepregnancy BMI (kg/m^2)	24.3 (22.3–28.9)	24.3 (22.3–28.0)	29.3 (24.1–36.2)
Maternal race/ethnicity			
White	90 (65)	49 (65)	41 (55)
Black	32 (13)	9 (12)	23 (31)
Hispanic	19 (16)	12 (16)	7 (9)
Other[b]	9 (7)	5 (7)	4 (5)
Maternal education			
High school or less	26 (15)	11 (15)	15 (20)
Some college/technical school	25 (12)	8 (11)	17 (23)
College or greater	96 (73)	54 (74)	42 (57)
Missing	3	2	1
Health insurance			
Private insurance/HMO	117 (86)	64 (86)	53 (74)
Self-pay or Medicaid/Mass Health	29 (14)	10 (14)	19 (26)
Missing	4	1	3
Parity			
Multiparous	81 (59)	45 (60)	36 (48)
Nulliparous	69 (41)	30 (40)	39 (52)
Smoking during pregnancy			
No	138 (93)	70 (93)	68 (91)
Yes	12 (7)	5 (7)	7 (9)
History of chronic hypertension			
No	132 (94)	71 (95)	61 (81)
Yes	18 (6)	4 (5)	14 (19)
Preeclampsia in a previous pregnancy			
No	139 (98)	74 (99)	65 (87)
Yes	11 (2)	1 (1)	10 (13)
Fetal sex			
Female	75 (48)	36 (48)	39 (52)
Male	75 (52)	39 (52)	36 (48)

Note: BMI, body mass index; HMO, health maintenance organization; IPW, inverse probability weight.
[a]Demographics presented for the total weighted population include IPWs to account for case–control selection and represent the weighted median (25th, 75th percentile) or the *n* (weighted %).
[b]Other includes Asian ($n=7$), Other ($n=2$) race and ethnicities.

hypertension (11% vs. 38%) and to have had preeclampsia in a previous pregnancy (9% vs. 24%) compared with those who developed early-onset preeclampsia.

The trajectories of angiogenic biomarkers were similar between cases and controls (Figure 1). For example, PlGF levels increase throughout pregnancy in both cases and controls, until late pregnancy, when levels begin to fall. However, the concentrations of these biomarkers differed between cases and controls (Table S4). Specifically, PlGF levels tended to be lower in cases (Visit 1: 19.3 vs. 20.0 ng/mL; Visit 2: 106 vs. 120 ng/mL; Visit 3: 280 vs. 444 ng/mL; and Visit 4: 165 vs. 366 ng/mL). Concentrations of sFLT-1 and the sFLT-1:PlGF ratio were lower among cases during early pregnancy and higher among cases later in pregnancy. Differences in the levels of angiogenic biomarkers were largest in magnitude when looking specifically at participants who developed early-onset preeclampsia compared with controls (Table S4). For example, concentrations of PlGF were lowest among early-onset preeclampsia cases (Visit 1: 18.2 ng/mL; Visit 2: 58.8 ng/mL; Visit 3: 163 ng/mL; and Visit 4: 111 ng/mL) compared with both late-onset cases (Visit 1:

Figure 1. Angiogenic biomarker concentrations across pregnancy by case status in the LIFECODES case–control study population, recruited from 2006 to 2008. (A) Maternal sFLT-1, (B) maternal PlGF, and (C) maternal sFLT-1:PlGF ratio. Individual data points are shown in circles for controls and triangles for cases. Lines (solid lines for controls, dashed lines for cases) and shading are locally estimated scatter smoothing and associated 95% confidence intervals. Data corresponding to this figure are in Table S4. Note: PlGF, placental growth factor; sFLT, soluble fms-like tyrosine kinase-1.

19.5 ng/mL; Visit 2: 110.9 ng/mL; Visit 3: 298 ng/mL; and Visit 4: 177 ng/mL) and controls (Visit 1: 20.0 ng/mL; Visit 2: 120 ng/mL; Visit 3: 444 ng/mL; and Visit 4: 366 ng/mL).

Concentrations of Plasma PFAS Biomarkers

In general, levels of PFAS in this cohort were lower than those reported in reproductive age females from the 2007–2008 cycle of NHANES (Table 2). For example, the GM of plasma PFOS concentrations in our cohort was 8.16 ng/mL compared with 10.7 ng/mL in NHANES. No differences in PFAS levels were noted when comparing controls with participants who developed any type of preeclampsia (Table S5). However, levels differed when preeclampsia cases were further categorized according to early- and late-onset. Specifically, PFOS levels were lowest among participants who developed early-onset preeclampsia and highest in those who developed late-onset preeclampsia (8.24 vs. 9.84 ng/mL). Similarly, levels of MeFOSAA, PFDA, and PFUnDA were lowest among participants who developed early-onset preeclampsia. Levels of PFNA were highest among individuals who developed late-onset preeclampsia (Table S5).

When compared across demographic characteristics, there were some notable trends in PFAS concentrations (Table S6). For example, white participants had the highest levels of PFHxS and PFOS compared with other race and ethnicities. Participants who smoked had lower levels of several PFAS, including PFDA and PFHxS. Multiparous participants had lower levels of PFOA, PFUnDA, PFHxS, and PFOS compared with nulliparous participants (Table S6).

Plasma PFAS Concentrations and Preeclampsia

Crude associations between PFAS and preeclampsia outcomes are shown in Table S7. After adjusting for maternal age, prepregnancy BMI, maternal race, maternal education, health insurance, parity, and gestational age at the time of sampling, the associations between most PFAS and any preeclampsia were elevated, although their CIs included the null (Figure 2; Table S8). For example, PFDA was marginally associated with preeclampsia: OR = 1.41 (95% CI: 1.00, 2.00). When separately examining preeclampsia subtypes, several associations were noted between PFAS and late-onset preeclampsia. Specifically, an IQR-increase in plasma concentrations of PFDA (OR = 1.64, 95% CI: 1.08, 2.47) and PFOS (OR = 1.60, 95% CI: 1.06, 2.43) was associated with higher odds of late-onset preeclampsia. On the other hand, the point estimates for associations between PFAS and early-onset preeclampsia were primarily below the null, although imprecise. For example, an IQR-increase in MeFOSAA was associated with an OR = 0.25 (95% CI: 0.06, 1.03) for early-onset preeclampsia. Models adjusting for chronic hypertension did not differ from the main results (Table S9). When restricted to nulliparous participants ($n = 69$), estimates for the association between PFAS and preeclampsia subtypes were much less precise. However, trends remained largely consistent (Table S10). For example, the point estimates for early-onset disease remained primarily below the null, whereas estimates for late-onset disease were still above the null. Specifically, an IQR-increase in PFDA was associated with an OR = 2.11 (95% CI: 0.99, 4.52) for late-onset preeclampsia.

When analyzing these data using Cox proportional hazards models, estimates for the association between PFAS and preeclampsia outcomes were largely similar (Table S11). When analyzing associations with late-onset preeclampsia, an IQR-increase in PFDA remained associated with elevated risk of disease (HR = 1.62, 95% CI: 1.25, 2.11). The estimate for an IQR-increase in PFOS (HR = 1.46, 95% CI: 0.91, 2.34) and late-onset preeclampsia remained above the null, although it was somewhat attenuated. Notably, the association between MeFOSAA and early-onset preeclampsia remained strongly below the null (HR = 0.33, 95% CI: 0.13, 0.85) and increased in precision.

Plasma PFAS Concentrations and Angiogenic Biomarkers

Crude associations between PFAS and angiogenic biomarkers are shown in Table S12. After adjusting for potential confounders, few associations between PFAS concentrations and angiogenic biomarkers were noted (Figure 3; Table S13). An IQR-increase in concentrations of PFHpA and PFOA was associated with lower PlGF concentrations, whereas PFUnDA was associated with higher PlGF at Visit 1. An IQR-increase in PFDA concentrations was also associated with lower PlGF concentrations at Visit 2, although associations were nonsignificant at other time points. There were some associations with sFLT-1, including persistent inverse associations between PFOS and sFLT-1 at Visits 2–4. Inverse associations between MeFOSAA and both sFLT-1 and the sFLT-1:PLGF ratio were also observed at Visits 2 and 3.

Discussion

In this case–control study, we examined the relationship between PFAS exposure, preeclampsia, and angiogenic biomarkers. To our knowledge, this study is the first to distinguish between

Table 2. Distribution of plasma PFAS concentrations (ng/mL) in the overall LIFECODES case–control study population, recruited from 2006 to 2008 ($n = 150$), and in women from the National Health and Examination Nutrition Survey (NHANES) 2007–2008.

					Percentiles				NHANES (2007–2008)
PFAS	Carbon chain length	LOD	*n* (%) >LOD	GM	25th	50th	75th	95th	GM
Perfluoroalkyl carboxylic acids									
PFHpA	7	0.1	144 (96)	0.43	0.21	0.63	0.76	1.00	<LOD
PFOA	8	0.5	149 (99)	2.76	2.12	2.92	3.37	4.80	3.55
PFNA	9	0.1	149 (99)	0.83	0.66	0.85	0.99	1.66	1.09
PFDA	10	0.1	146 (97)	0.31	0.25	0.30	0.42	0.68	0.27
PFUnDA	11	0.1	127 (85)	0.23	0.15	0.23	0.38	0.55	<LOD
Perfluoroalkyl sulfonates									
PFHxS	6	0.1	149 (99)	0.94	0.53	0.88	1.36	2.94	1.46
PFOS	8	0.1	149 (99)	8.16	5.77	8.45	10.7	18.1	10.7
Perfluoroalkyl sulfonamides									
MeFOSAA	8	0.1	125 (83)	0.19	0.11	0.17	0.27	0.71	0.30
PFOSA	8	0.1	5 (3)	—	—	—	—	—	<LOD

Note: Descriptive statistics for plasma PFAS concentrations in the overall study population include IPWs to account for case–control selection. —, Not applicable; GM, geometric mean; IPW, inverse probability weight; LOD, limit of detection; MeFOSAA, 2-(*N*-methyl-perfluorooctane sulfonamido) acetic acid; PDFA, perfluorodecanoic acid; PFAS, per- and polyfluoroalkyl substances; PFHpA, perfluoroheptanoic acid; PFHxS, perfluorohexane sulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonic acid; PFOSA, perfluorooctane sulfonamide; PFUnDA, perfluoroundecanoic acid.

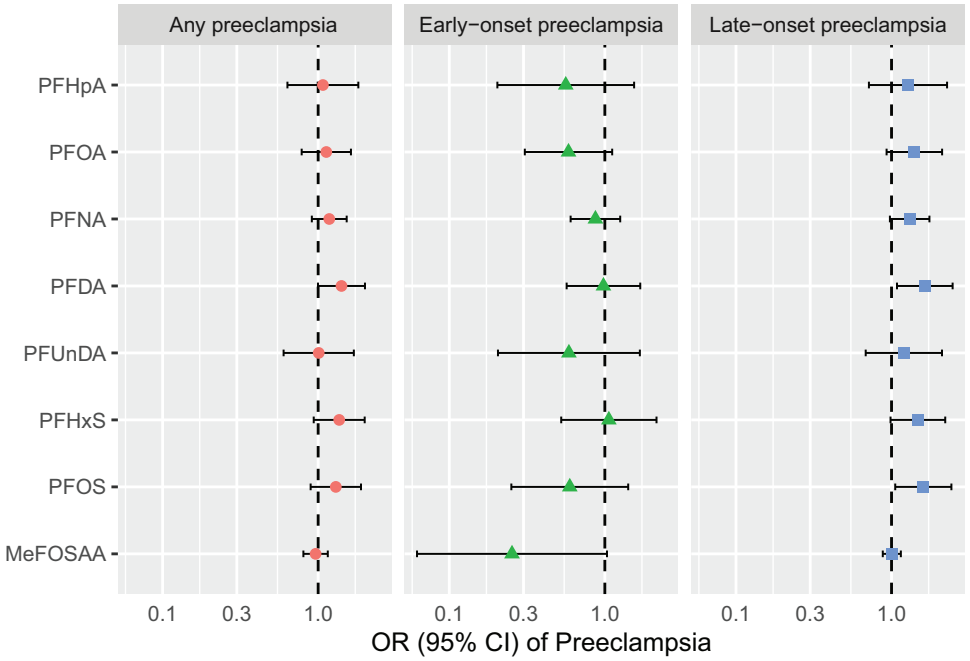

Figure 2. Adjusted ORs (95% CIs) of preeclampsia outcomes associated with an IQR-increase in PFAS in the LIFECODES case–control study population recruited from 2006 to 2008. Point estimates for (A) any preeclampsia (circles), (B) early-onset preeclampsia (triangles), and (C) late-onset preeclampsia (squares) are flanked by lines that indicate the 95% CI. Estimates are from logistic regression models adjusted for maternal age (in years), prepregnancy BMI (in kg/m^2), maternal race (non-Black/Black), parity (nulliparous/parous), maternal education (high school or less/some college or technical school/college or greater), health insurance (private insurance/self-pay or public insurance), and gestational age at time of sample collection (in weeks). Case $n = 71$ ($n = 19$ early-onset and $n = 52$ late-onset), control $n = 73$. Data corresponding to this figure are in Table S8. Note: BMI, body mass index; CI, confidence interval; IQR, interquartile range; MeFOSAA, 2-(*N*-methyl-perfluorooctane sulfonamido) acetic acid; OR, odds ratio; PDFA, perfluorodecanoic acid; PFAS, per- and polyfluoroalkyl substances; PFHpA, perfluoroheptanoic acid; PFHxS, perfluorohexane sulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonic acid; PFUnDA, perfluoroundecanoic acid.

preeclampsia subtypes, which are believed to have distinct etiologies (Raymond and Peterson 2011; Roberts et al. 2021; von Dadelszen et al. 2003) and may therefore have different associations with environmental exposures, such as PFAS. We observed that plasma concentrations of both PFDA and PFOS were associated with higher odds of late-onset preeclampsia in our main analysis. The associations between PFAS and early-onset preeclampsia were primarily below the null, although imprecise. Last, we examined whether PFAS were associated with changes in circulating angiogenic biomarkers. Few significant associations were noted, and point estimates were consistently below the null when examining PFAS in relation to angiogenic biomarker concentrations.

Participants in the present study had lower levels of many PFAS compared with concentrations of PFAS reported in women of reproductive age in NHANES (cycle years: 2007–2008). However, concentrations in this study are within the range of PFAS measurements reported in other studies on this topic. For example, in a case–control study nested in the Norwegian Mother and Baby (MoBA) study ($n = 976$), maternal plasma concentrations of PFOA (median = 2.78 ng/mL) were similar to those reported in this cohort (PFOA median = 2.92 ng/mL), although PFOS concentrations were somewhat higher (MoBA median = 12.87 ng/mL vs. LIFECODES median = 8.45 ng/mL) (Starling et al. 2014). Other cohorts, such as the prospective Maternal-Infant Research on Environmental Chemicals (MIREC, $n = 1{,}739$) study and the Swedish Environmental Longitudinal, Mother and Child, Asthma and Allergy (SELMA, $n = 1{,}773$) pregnancy cohort, have reported lower concentrations of PFOA and PFOS (Borghese et al. 2020; Wikström et al. 2019). Because measures of some legacy PFAS in the general population are declining over time (Göckener et al. 2020; Seo et al. 2018), the concentrations measured in the present study may still be relevant to assessing the ongoing risks of these exposures.

When examining preeclampsia subtypes, we observed that PFDA and PFOS were associated with higher odds of late-onset preeclampsia. On the other hand, associations between PFAS and early-onset preeclampsia were below the null, although imprecise. Notably, work from the C8 Health Study ($n = 5{,}262$) and the SELMA study have previously reported associations between higher PFOS exposure and preeclampsia incidence (Stein et al. 2009; Wikström et al. 2019). Nevertheless, findings across studies have been mixed and many, including a previous study from the MoBA cohort, have reported largely null associations between PFAS and preeclampsia diagnosis (Huo et al. 2020; Rylander et al. 2020; Starling et al. 2014). One possible reason for this heterogeneity is that although preeclampsia is commonly studied as a single disorder, the disease is highly heterogeneous, with multiple proposed pathophysiologic pathways and subtypes (Roberts et al. 2021; Staff 2019). For example, early- and late-onset preeclampsia have distinct, although overlapping, risk factors. Specifically, chronic hypertension or previous preeclampsia diagnosis is a stronger risk factor for early-onset preeclampsia, whereas diabetes mellitus and nulliparity are more strongly associated with late-onset preeclampsia according to previous studies (Lisonkova and Joseph 2013; You et al. 2018). Such risk factors may differ between cohorts being sampled from different source populations based on geographic location or other factors. Because mechanistic pathways may differ between preeclampsia subtypes (Staff 2019; von Dadelszen et al. 2003), incorporating information about disease phenotypes may facilitate the identification of exposures that could contribute to specific subtypes of the disease.

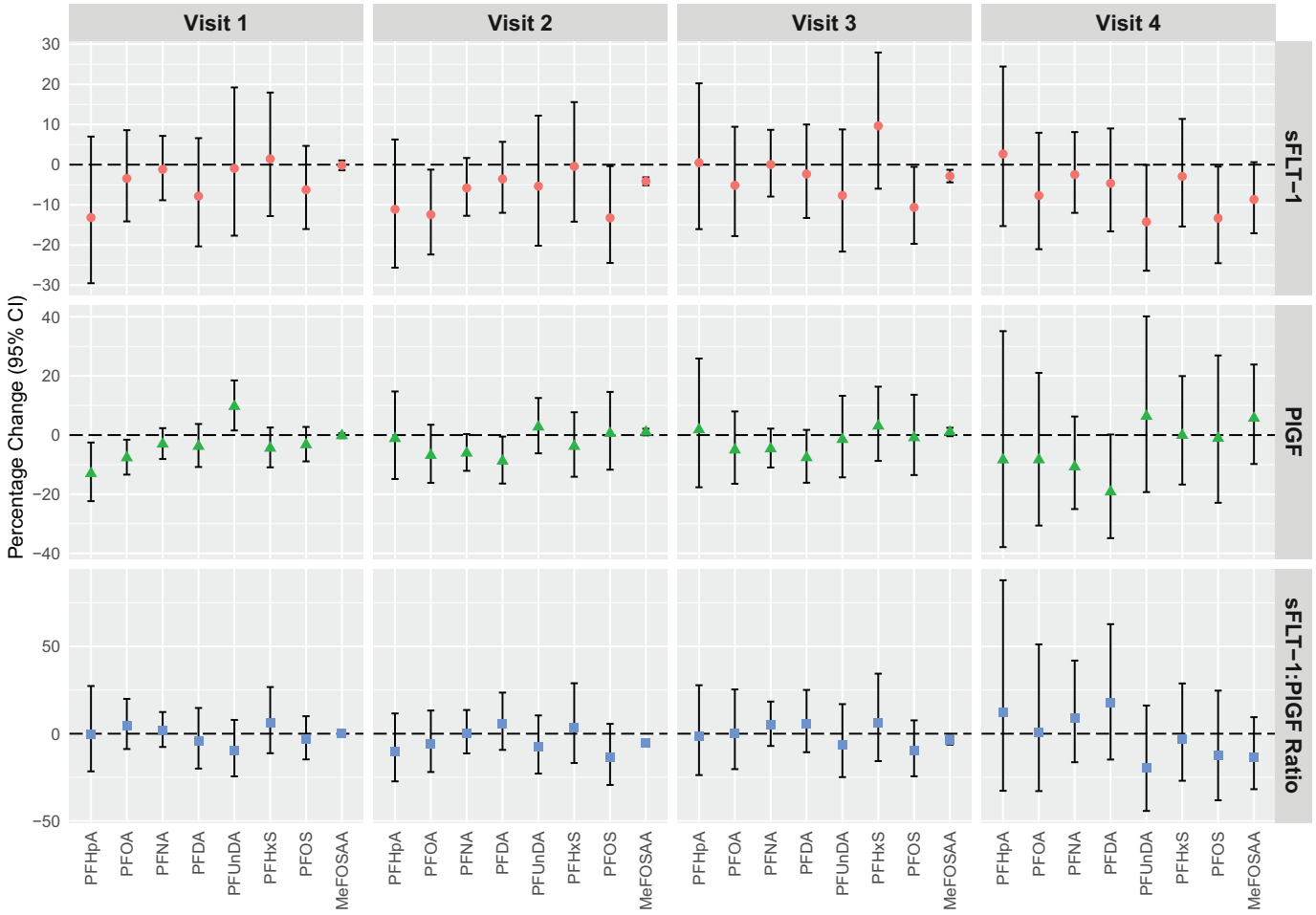

Figure 3. Adjusted percentage change (95% CI) in angiogenic biomarkers associated with an IQR-increase in PFAS in the LIFECODES case–control study population, recruited from 2006 to 2008. Estimated percentage changes are displayed with circles (sFLT-1), triangles (PlGF) and squares (sFLT-1:PlGF ratio) and are flanked by lines that indicate the 95% CI. Estimates are from linear regression models adjusted for maternal age (in years), prepregnancy BMI (in kg/m^2), maternal race (non-Black/Black), parity (nulliparous/parous), maternal education (high school or less/some college or technical school/college or greater), health insurance (private insurance/self-pay or public insurance), and gestational age at time of sample collection (in weeks). Visit 1 $n = 138$, Visit 2 $n = 128$, Visit 3 $n = 126$, and Visit 4 $n = 124$. Data corresponding to this figure are in Table S13. Note: BMI, body mass index; CI, confidence interval; IQR, interquartile range; MeFOSAA, 2-(*N*-methyl-perfluorooctane sulfonamido) acetic acid; PDFA, perfluorodecanoic acid; PFAS, per- and polyfluoroalkyl substances; PFHpA, perfluoroheptanoic acid; PFHxS, perfluorohexane sulfonic acid; PFNA, perfluorononanoic acid; PFOA, perfluorooctanoic acid; PFOS, perfluorooctane sulfonic acid; PFUnDA, perfluoroundecanoic acid; PlGF, placental growth factor; sFLT-1, soluble fms-like tyrosine kinase 1.

Understanding our findings warrants greater consideration of the different characteristics of early- and late-onset preeclampsia. Classically, all preeclampsia has been believed to be initiated by a failure of placental trophoblastic cells to adequately invade into maternal decidua and replace the endothelial lining of the uterine spiral arteries, which is a process that occurs early during pregnancy (Pijnenborg et al. 2006; Steegers et al. 2010; Zhou et al. 1997). However, this etiology is more strongly associated with the early-onset phenotype of preeclampsia (Raymond and Peterson 2011). On the other hand, late-onset preeclampsia often appears to occur even when placental development is sufficient. Instead, this phenotype is characterized by placental underperfusion, where the placenta has outgrown its supply or its supply is limited (Redman and Staff 2015; Roberts and Hubel 2009). It has been proposed that this underperfusion may be the result of excessive trophoblast senescence (Cox and Redman 2017) or other intrinsic maternal factors, such as obesity, which may contribute to higher levels of placental stress (Staff 2019; Steegers et al. 2010). Thus, although both phenotypes involve suboptimal placental perfusion, the two are distinct in their underlying pathophysiology. Our findings in the present study suggest that PFAS exposure may be more closely related to late-onset preeclampsia and, therefore, to mechanisms aside from aberrant placentation. However, it should be noted that existing toxicologic evidence indicates that PFAS may alter processes that are implicated in early-onset disease, including trophoblast migration and invasion (Szilagyi et al. 2020) and angiogenic processes (Pham et al. 2020; Poteser et al. 2020). Exposure to PFAS has also been shown to alter placental weight, disrupt its labyrinth structure, and induce congestion and clot formation (Blake et al. 2020), as well as apoptosis (Jiang et al. 2020), in *in vivo* studies. In light of this mechanistic evidence, further consideration of the relationship between PFAS and preeclampsia subtypes is warranted. In addition, most mechanistic studies on the effects of PFAS on placental development and function have focused on a small number of PFAS (e.g., PFOS and PFOA) and further research into a greater number of substances would foster an increased understanding of the results observed in the present study and others.

It is possible that our estimates for the association between PFAS and preeclampsia could be influenced by reverse causation. A range of human studies, including cross-sectional and prospective studies, have demonstrated that the distribution and excretion

of PFAS is sensitive to changes in kidney function (Blake et al. 2018; Jain and Ducatman 2019; Sagiv et al. 2015), especially albumin excretion given that albumin is the primary carrier protein of PFAS in the blood (Forsthuber et al. 2020). Specifically, higher urinary albumin and lower serum albumin levels have been shown to correspond to lower concentrations of plasma PFAS (Jain and Ducatman 2019; Sagiv et al. 2015). Although we measured PFAS concentrations well before the onset of preeclampsia, urinary albumin levels have been observed to increase months before the disease becomes clinically apparent (Babu et al. 2015; Baweja et al. 2011; Ekbom et al. 2000; Jensen et al. 2010; Shaarawy and Salem 2001). We expect that this would disproportionately impact our estimates for early-onset preeclampsia (samples collected 12–25 wk prior to disease onset) compared with late-onset cases (samples collected 21–32 wk prior to disease onset). This may be reflected in our crude analysis of PFAS concentrations, where concentrations of several PFAS were lowest in women who developed early-onset preeclampsia compared with both controls and late-onset preeclampsia cases. Thus, it is possible that plasma PFAS levels in women who developed early-onset preeclampsia were lower, in part, because of already-changing patterns of albumin excretion.

In general, associations between PFAS and angiogenic biomarkers were null. These biomarkers are better predictors of early-onset preeclampsia (Levine et al. 2004; Pinheiro et al. 2014). Thus, the lack of robust associations with the angiogenic biomarkers is consistent with our observation that PFAS were primarily associated with late-onset preeclampsia. Nevertheless, we found that an increase in PFDA was associated with lower PlGF concentrations at Visit 2 and was weakly associated with lower PlGF concentrations at Visit 4. Given that higher circulating concentrations of PlGF negatively predict preeclampsia risk (Agrawal et al. 2019), this suggests that greater exposure to PFDA is associated with increased risk of preeclampsia and aligns with our observation that PFDA was associated with higher odds of late-onset preeclampsia. On the other hand, an IQR-increase in MeFOSAA was associated with lower sFLT-1 and a lower sFLT-1:PlGF ratio at Visits 2 and 3. Notably, PFOS was also associated with lower sFLT-1 concentrations at Visits 2 through 4. Higher sFLT-1 and a higher sFLT-1:PlGF ratio are typically thought to be indicative of greater preeclampsia risk (Agrawal et al. 2019; Allen et al. 2014; Veisani et al. 2019). However, in the present study, sFLT-1 concentrations and the sFLT-1:PlGF ratio were not consistently higher among women who developed preeclampsia until late pregnancy (i.e., Visit 4). Taken together, these findings provide weak support for the adverse influence of PFAS on indicators of placental angiogenesis and, instead, suggest that PFAS may influence preeclampsia via other mechanisms, such as maternal inflammatory or metabolic dysregulation.

This study is not without limitations. We used a case–control population sampled from patients at Brigham and Women's Hospital. This study center serves as a tertiary care clinic and receives higher-risk pregnancies, which may limit the generalizability of these results. In addition, the study sample was small to moderate in size. However, the case–control sampling scheme resulted in having many preeclampsia cases ($n=75$) relative to several other large cohort studies on this topic (Borghese et al. 2020; Huo et al. 2020; Wikström et al. 2019). Also in contrast to other studies on this topic, the present study sampled women who were multiparous, as well as those who had preexisting chronic hypertension. Many previous studies have chosen to exclude these women because the confounding structure of the PFAS–preeclampsia relationship may differ depending on parity (Starling et al. 2014) and because chronic hypertension is known to make the diagnosis of preeclampsia more difficult and potentially less reliable (ACOG 2013). Although we performed an analysis restricted to nulliparous women, the small number of nulliparous women in this cohort ($n=69$) limited our ability to fully examine this issue. Likewise, the diagnosis of preeclampsia in this study was rigorously validated by at least two Maternal–Fetal Medicine physicians and sensitivity analyses adjusting for chronic hypertension did not change the conclusions of our main results. Last, we were missing information on several factors related to reproductive characteristics that could have an impact on both PFAS concentrations and pregnancy outcomes, including interpregnancy interval or new paternity, and menstrual characteristics (Singer et al. 2018).

The present study also has several strengths. First, as previously mentioned, the rigorous validation of preeclampsia cases allowed for the determination of early- and late-onset preeclampsia subtypes. Preeclampsia is heterogenous. Although it is diagnosed based on a shared set of symptoms (i.e., hypertension and proteinuria), the pathophysiological diversity in the disorder supports separating it into distinct subcategories (Raymond and Peterson 2011; van der Merwe et al. 2010). In this study, we found that associations could differ depending on the preeclampsia subtype, underscoring the importance of being able to incorporate this information into future studies of preeclampsia. In addition, having estimates of disease onset allowed for secondary analyses of these data using Cox proportional hazard models, which more adequately accounted for preterm birth as a competing risk (Weinberg et al. 2017). This may be relevant in the study of PFAS and preeclampsia given that PFAS have also been associated with a higher risk of preterm birth (Meng et al. 2018; Waterfield et al. 2020). Last, this study measured PFAS exposure using maternal plasma collected in early pregnancy. It is known that kidney function, which changes across pregnancy, impacts the distribution and excretion of PFAS (Sagiv et al. 2018). Thus, having an early measure of PFAS exposure minimizes the potential that our exposure measures are biased from these effects.

Conclusions

We observed that several PFAS were associated with increased odds of late-onset preeclampsia. Associations with early-onset preeclampsia were primarily below the null, although findings were imprecise given the small number of cases. One possible explanation for inconsistency in the current literature on PFAS and preeclampsia may, in part, be due to not considering the heterogeneity of this disease.

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References

ACOG (American College of Obstetrics and Gynecologists). 2013. Hypertension in pregnancy. Report of the American College of Obstetricians and Gynecologists' task force on hypertension in pregnancy. Obstet Gynecol 122(5):1122–1131, PMID: 24150027, https://doi.org/10.1097/01.AOG.0000437382.03963.88.

ACOG. 2017. Committee Opinion no 700: methods for estimating the due date. Obstet Gynecol 129(5):e150–e154, PMID: 28426621, https://doi.org/10.1097/AOG.0000000000002046.

ACOG. 2020. Gestational hypertension and preeclampsia: ACOG Practice Bulletin Summary, number 222. Obstet Gynecol 135(6):e237–e260, PMID: 32443077, https://doi.org/10.1097/AOG.0000000000003892.

Agrawal S, Shinar S, Cerdeira AS, Redman C, Vatish M. 2019. Predictive performance of PlGF (placental growth factor) for screening preeclampsia in asymptomatic women: a systematic review and meta-analysis. Hypertension 74(5):1124–1135, PMID: 31522621, https://doi.org/10.1161/HYPERTENSIONAHA.119.13360.

Allen RE, Rogozinska E, Cleverly K, Aquilina J, Thangaratinam S. 2014. Abnormal blood biomarkers in early pregnancy are associated with preeclampsia: a meta-analysis. Eur J Obstet Gynecol Reprod Biol 182:194–201, PMID: 25305662, https://doi.org/10.1016/j.ejogrb.2014.09.027.

Ananth CV, Keyes KM, Wapner RJ. 2013. Pre-eclampsia rates in the United States, 1980–2010: age-period-cohort analysis. BMJ 347:f6564, PMID: 24201165, https://doi.org/10.1136/bmj.f6564.

Ashley-Martin J, Dodds L, Arbuckle TE, Morisset A-S, Fisher M, Bouchard MF, et al. 2016. Maternal and neonatal levels of perfluoroalkyl substances in relation to gestational weight gain. Int J Environ Res Public Health 13(1):146, PMID: 26805861, https://doi.org/10.3390/ijerph13010146.

ATSDR (Agency for Toxic Substances and Disease Registry). 2021. *Toxicological Profile for Perfluoroalkyls*. Atlanta, GA, ATSDR. https://www.atsdr.cdc.gov/ToxProfiles/tp200.pdf [accessed 27 September 2021].

Babu RP, Christy A, Hegde A, Manjrekar P, Joseph M. 2015. Microalbuminuria—a better marker in hypertensive disorders of pregnancy. Saudi J Kidney Dis Transpl 26(5):941–946, PMID: 26354565, https://doi.org/10.4103/1319-2442.164574.

Baweja S, Kent A, Masterson R, Roberts S, McMahon LP. 2011. Prediction of pre-eclampsia in early pregnancy by estimating the spot urinary albumin: creatinine ratio using high-performance liquid chromatography. BJOG 118(9):1126–1132, PMID: 21481153, https://doi.org/10.1111/j.1471-0528.2011.02960.x.

Bellamy L, Casas J-P, Hingorani AD, Williams DJ. 2007. Pre-eclampsia and risk of cardiovascular disease and cancer in later life: systematic review and meta-analysis. BMJ 335(7627):974, PMID: 17975258, https://doi.org/10.1136/bmj.39335.385301.BE.

Blake BE, Cope HA, Hall SM, Keys RD, Mahler BW, McCord J, et al. 2020. Evaluation of maternal, embryo, and placental effects in CD-1 mice following gestational exposure to perfluorooctanoic acid (PFOA) or hexafluoropropylene oxide dimer acid (HFPO-DA or GenX). Environ Health Perspect 128(2):27006, PMID: 32074459, https://doi.org/10.1289/EHP6233.

Blake BE, Fenton SE. 2020. Early life exposure to per- and polyfluoroalkyl substances (PFAS) and latent health outcomes: a review including the placenta as a target tissue and possible driver of peri- and postnatal effects. Toxicology 443:152565, PMID: 32861749, https://doi.org/10.1016/j.tox.2020.152565.

Blake BE, Pinney SM, Hines EP, Fenton SE, Ferguson KK. 2018. Associations between longitudinal serum perfluoroalkyl substance (PFAS) levels and measures of thyroid hormone, kidney function, and body mass index in the Fernald Community Cohort. Environ Pollut 242(pt A):894–904, PMID: 30373035, https://doi.org/10.1016/j.envpol.2018.07.042.

Borghese MM, Walker M, Helewa ME, Fraser WD, Arbuckle TE. 2020. Association of perfluoroalkyl substances with gestational hypertension and preeclampsia in the MIREC study. Environ Int 141:105789, PMID: 32408216, https://doi.org/10.1016/j.envint.2020.105789.

Calafat AM, Kato K, Hubbard K, Jia T, Botelho JC, Wong L-Y. 2019. Legacy and alternative *per-* and polyfluoroalkyl substances in the U.S. general population: paired serum-urine data from the 2013–2014 National Health and Nutrition Examination Survey. Environ Int 131:105048, PMID: 31376596, https://doi.org/10.1016/j.envint.2019.105048.

CDC (Centers for Disease Control and Prevention). 2019. *Fourth National Report on Human Exposure to Environmental Chemicals—Updated Tables, January 2019, Volume One*. https://www.cdc.gov/exposurereport/index.html [accessed 27 September 2021].

Cox LS, Redman C. 2017. The role of cellular senescence in ageing of the placenta. Placenta 52:139–145, PMID: 28131318, https://doi.org/10.1016/j.placenta.2017.01.116.

Ekbom P, Damm P, Nørgaard K, Clausen P, Feldt-Rasmussen U, Feldt-Rasmussen B, et al. 2000. Urinary albumin excretion and 24-hour blood pressure as predictors of pre-eclampsia in type I diabetes. Diabetologia 43(7):927–931, PMID: 10952467, https://doi.org/10.1007/s001250051471.

Forsthuber M, Kaiser AM, Granitzer S, Hassl I, Hengstschläger M, Stangl H, et al. 2020. Albumin is the major carrier protein for PFOS, PFOA, PFHxS, PFNA and PFDA in human plasma. Environ Int 137:105324, PMID: 32109724, https://doi.org/10.1016/j.envint.2019.105324.

Göckener B, Weber T, Rüdel H, Bücking M, Kolossa-Gehring M. 2020. Human biomonitoring of per- and polyfluoroalkyl substances in German blood plasma samples from 1982 to 2019. Environ Int 145:106123, PMID: 32949877, https://doi.org/10.1016/j.envint.2020.106123.

Honigberg MC, Cantonwine DE, Thomas AM, Lim K-H, Parry SI, McElrath TF. 2016. Analysis of changes in maternal circulating angiogenic factors throughout pregnancy for the prediction of preeclampsia. J Perinatol 36(3):172–177, PMID: 26583938, https://doi.org/10.1038/jp.2015.170.

Hornung RW, Reed LD. 1990. Estimation of average concentration in the presence of nondetectable values. Appl Occup Environ Hyg 5(1):46–51, https://doi.org/10.1080/1047322X.1990.10389587.

Huang R, Chen Q, Zhang L, Luo K, Chen L, Zhao S, et al. 2019. Prenatal exposure to perfluoroalkyl and polyfluoroalkyl substances and the risk of hypertensive disorders of pregnancy. Environ Health 18(1):5, PMID: 30626391, https://doi.org/10.1186/s12940-018-0445-3.

Huo X, Huang R, Gan Y, Luo K, Aimuzi R, Nian M, et al. 2020. Perfluoroalkyl substances in early pregnancy and risk of hypertensive disorders of pregnancy: a prospective cohort study. Environ Int 138:105656, PMID: 32222612, https://doi.org/10.1016/j.envint.2020.105656.

ITRC (Interstate Technology and Regulatory Council). 2020a. History and use of per- and polyfluoroalkyl substances (PFAS). [Fact sheet.] https://pfas-1.itrcweb.org/fact_sheets_page/PFAS_Fact_Sheet_History_and_Use_April2020.pdf [accessed 27 September 2021].

ITRC. 2020b. Naming conventions and physical and chemical properties of per- and polyfluoroalkyl substances (PFAS). [Fact sheet.] https://pfas-1.itrcweb.org/fact_sheets_page/PFAS_Fact_Sheet_Naming_Conventions_April2020.pdf [accessed 27 September 2021].

Jain RB, Ducatman A. 2019. Perfluoroalkyl acids serum concentrations and their relationship to biomarkers of renal failure: serum and urine albumin, creatinine, and albumin creatinine ratios across the spectrum of glomerular function among US adults. Environ Res 174:143–151, PMID: 31077989, https://doi.org/10.1016/j.envres.2019.04.034.

Jensen DM, Damm P, Ovesen P, Mølsted-Pedersen L, Beck-Nielsen H, Westergaard JG, et al. 2010. Microalbuminuria, preeclampsia, and preterm delivery in pregnant women with type 1 diabetes: results from a nationwide Danish study. Diabetes Care 33(1):90–94, PMID: 19846800, https://doi.org/10.2337/dc09-1219.

Jiang W, Deng Y, Song Z, Xie Y, Gong L, Chen Y, et al. 2020. Gestational perfluorooctanoic acid exposure inhibits placental development by dysregulation of labyrinth vessels and uNK cells and apoptosis in mice. Front Physiol 11:51, PMID: 32116763, https://doi.org/10.3389/fphys.2020.00051.

Kenneth L, Hall DR, Gebhardt S, Grové D. 2010. Late onset preeclampsia is not an innocuous condition. Hypertens Pregnancy 29(3):262–270, PMID: 20670151, https://doi.org/10.3109/10641950902777697.

Kuklenyik Z, Needham LL, Calafat AM. 2005. Measurement of 18 perfluorinated organic acids and amides in human serum using on-line solid-phase extraction. Anal Chem 77(18):6085–6091, PMID: 16159145, https://doi.org/10.1021/ac050671l.

Levine RJ, Maynard SE, Qian C, Lim K-H, England LJ, Yu KF, et al. 2004. Circulating angiogenic factors and the risk of preeclampsia. N Engl J Med 350(7):672–683, PMID: 14764923, https://doi.org/10.1056/NEJMoa031884.

Lisonkova S, Joseph KS. 2013. Incidence of preeclampsia: risk factors and outcomes associated with early- versus late-onset disease. Am J Obstet Gynecol 209(6):544.e1–544.e12, PMID: 23973398, https://doi.org/10.1016/j.ajog.2013.08.019.

Maynard SE, Min J-Y, Merchan J, Lim K-H, Li J, Mondal S, et al. 2003. Excess placental soluble fms-like tyrosine kinase 1 (sFlt1) may contribute to endothelial dysfunction, hypertension, and proteinuria in preeclampsia. J Clin Invest 111(5):649–658, PMID: 12618519, https://doi.org/10.1172/JCI17189.

McElrath TF, Lim K-H, Pare E, Rich-Edwards J, Pucci D, Troisi R, et al. 2012. Longitudinal evaluation of predictive value for preeclampsia of circulating angiogenic factors through pregnancy. Am J Obstet Gynecol 207(5):407.e1–407.e7, PMID: 22981320, https://doi.org/10.1016/j.ajog.2012.08.010.

Meng Q, Inoue K, Ritz B, Olsen J, Liew Z. 2018. Prenatal exposure to perfluoroalkyl substances and birth outcomes; an updated analysis from the Danish National Birth Cohort. Int J Environ Res Public Health 15(9):1832, PMID: 30149566, https://doi.org/10.3390/ijerph15091832.

Mitro SD, Sagiv SK, Rifas-Shiman SL, Calafat AM, Fleisch AF, Jaacks LM, et al. 2020. Per- and polyfluoroalkyl substance exposure, gestational weight gain, and postpartum weight changes in Project Viva. Obesity (Silver Spring) 28(10):1984–1992, PMID: 32959518, https://doi.org/10.1002/oby.22933.

Mongraw-Chaffin ML, Cirillo PM, Cohn BA. 2010. Preeclampsia and cardiovascular disease death: prospective evidence from the Child Health and Development Studies cohort.. Hypertension 56(1):166–171, PMID: 20516394, https://doi.org/10.1161/HYPERTENSIONAHA.110.150078.

Pham A, Zhang J, Feng L. 2020. Exposure to perfluorobutane sulfonate and perfluorooctanesulfonic acid disrupts the production of angiogenesis factors and stress responses in human placental syncytiotrophoblast. Reprod Toxicol 98:269–277, PMID: 33144174, https://doi.org/10.1016/j.reprotox.2020.10.013.

Pijnenborg R, Vercruysse L, Hanssens M. 2006. The uterine spiral arteries in human pregnancy: facts and controversies. Placenta 27(9–10):939–958, PMID: 16490251, https://doi.org/10.1016/j.placenta.2005.12.006.

Pinheiro CC, Rayol P, Gozzani L, dos Reis LM, Zampieri G, Dias CB, et al. 2014. The relationship of angiogenic factors to maternal and neonatal manifestations of early-onset and late-onset preeclampsia. Prenat Diagn 34(11):1084–1092, PMID: 24916790, https://doi.org/10.1002/pd.4432.

Poteser M, Hutter H-P, Moshammer H, Weitensfelder L. 2020. Perfluoroctanoic acid (PFOA) enhances NOTCH-signaling in an angiogenesis model of placental trophoblast cells. Int J Hyg Environ Health 229:113566, PMID: 32485599, https://doi.org/10.1016/j.ijheh.2020.113566.

Raymond D, Peterson E. 2011. A critical review of early-onset and late-onset preeclampsia. Obstet Gynecol Surv 66(8):497–506, PMID: 22018452, https://doi.org/10.1097/OGX.0b013e3182331028.

Redman CWG, Staff AC. 2015. Preeclampsia, biomarkers, syncytiotrophoblast stress, and placental capacity. Am J Obstet Gynecol 213: (4 suppl):S9.e1–S9.e11, PMID: 26428507, https://doi.org/10.1016/j.ajog.2015.08.003.

Roberts JM, Hubel CA. 2009. The two stage model of preeclampsia: variations on the theme. Placenta 30(suppl A):S32–S37, PMID: 19070896, https://doi.org/10.1016/j.placenta.2008.11.009.

Roberts JM, Rich-Edwards JW, McElrath TF, Garmire L, Myatt L, Global Pregnancy Collaboration. 2021. Subtypes of preeclampsia: recognition and determining clinical usefulness. Hypertension 77(5):1430–1441, PMID: 33775113, https://doi.org/10.1161/HYPERTENSIONAHA.120.14781.

Rylander L, Lindh CH, Hansson SR, Broberg K, Källén K. 2020. Per- and polyfluoroalkyl substances in early pregnancy and risk for preeclampsia: a case–control study in southern Sweden. Toxics 8(2):43, PMID: 32560030, https://doi.org/10.3390/toxics8020043.

Sagiv SK, Rifas-Shiman SL, Fleisch AF, Webster TF, Calafat AM, Ye X, et al. 2018. Early-pregnancy plasma concentrations of perfluoroalkyl substances and birth outcomes in Project Viva: confounded by pregnancy hemodynamics? Am J Epidemiol 187(4):793–802, PMID: 29155920, https://doi.org/10.1093/aje/kwx332.

Sagiv SK, Rifas-Shiman SL, Webster TF, Mora AM, Harris MH, Calafat AM, et al. 2015. Sociodemographic and perinatal predictors of early pregnancy per- and polyfluoroalkyl substance (PFAS) concentrations. Environ Sci Technol 49(19):11849–11858, PMID: 26333069, https://doi.org/10.1021/acs.est.5b02489.

Savitz DA, Stein CR, Bartell SM, Elston B, Gong J, Shin H-M, et al. 2012. Perfluorooctanoic acid exposure and pregnancy outcome in a highly exposed community. Epidemiology 23(3):386–392, PMID: 22370857, https://doi.org/10.1097/EDE.0b013e31824cb93b.

Seo S-H, Son M-H, Choi S-D, Lee D-H, Chang Y-S. 2018. Influence of exposure to perfluoroalkyl substances (PFASs) on the Korean general population: 10-year trend and health effects. Environ Int 113:149–161, PMID: 29425899, https://doi.org/10.1016/j.envint.2018.01.025.

Shaarawy M, Salem ME. 2001. The clinical value of microtransferrinuria and microalbuminuria in the prediction of pre-eclampsia. Clin Chem Lab Med 39(1):29–34, PMID: 11256797, https://doi.org/10.1515/CCLM.2001.008.

Singer AB, Whitworth KW, Haug LS, Sabaredzovic A, Impinen A, Papadopoulou E, et al. 2018. Menstrual cycle characteristics as determinants of plasma concentrations of perfluoroalkyl substances (PFASs) in the Norwegian Mother and Child Cohort (MOBA study). Environ Res 166:78–85, PMID: 29879567, https://doi.org/10.1016/j.envres.2018.05.019.

Staff AC. 2019. The two-stage placental model of preeclampsia: an update. J Reprod Immunol 134–135:1–10, PMID: 31301487, https://doi.org/10.1016/j.jri.2019.07.004.

Starling AP, Engel SM, Richardson DB, Baird DD, Haug LS, Stuebe AM, et al. 2014. Perfluoroalkyl substances during pregnancy and validated preeclampsia among nulliparous women in the Norwegian Mother and Child Cohort study. Am J Epidemiol 179(7):824–833, PMID: 24557813, https://doi.org/10.1093/aje/kwt432.

Steegers EAP, von Dadelszen P, Duvekot JJ, Pijnenborg R. 2010. Pre-eclampsia. Lancet 376(9741):631–644, PMID: 20598363, https://doi.org/10.1016/S0140-6736(10)60279-6.

Stein CR, Savitz DA, Dougan M. 2009. Serum levels of perfluorooctanoic acid and perfluorooctane sulfonate and pregnancy outcome. Am J Epidemiol 170(7):837–846, PMID: 19692329, https://doi.org/10.1093/aje/kwp212.

Sunderland EM, Hu XC, Dassuncao C, Tokranov AK, Wagner CC, Allen JG. 2019. A review of the pathways of human exposure to poly-and perfluoroalkyl substances (PFASs) and present understanding of health effects. J Expo Sci Environ Epidemiol 29(2):131–147, PMID: 30470793, https://doi.org/10.1038/s41370-018-0094-1.

Szilagyi JT, Freedman AN, Kepper SL, Keshava AM, Bangma JT, Fry RC. 2020. Per- and polyfluoroalkyl substances (PFAS) differentially inhibit placental trophoblast migration and invasion *in vitro*. Toxicol Sci 175(2):210–219, PMID: 32219433, https://doi.org/10.1093/toxsci/kfaa043.

Timmermann CAG, Jensen KJ, Nielsen F, Budtz-Jørgensen E, van der Klis F, Benn CS, et al. 2020. Serum perfluoroalkyl substances, vaccine responses, and morbidity in a cohort of Guinea-Bissau children. Environ Health Perspect 128(8):087002, PMID: 32772733, https://doi.org/10.1289/EHP6517.

van der Merwe JL, Hall DR, Wright C, Schubert P, Grové D. 2010. Are early and late preeclampsia distinct subclasses of the disease—what does the placenta reveal? Hypertens Pregnancy 29(4):457–467, PMID: 20701467, https://doi.org/10.3109/10641950903572282.

Veisani Y, Jenabi E, Delpisheh A, Khazaei S. 2019. Angiogenic factors and the risk of preeclampsia: a systematic review and meta-analysis. Int J Repro Biomed 17(1):1–10, PMID: 31435580, https://doi.org/10.18502/ijrm.v17i1.3815.

von Dadelszen P, Magee LA, Roberts JM. 2003. Subclassification of preeclampsia. Hypertens Pregnancy 22(2):143–148, PMID: 12908998, https://doi.org/10.1081/PRG-120021060.

Waterfield G, Rogers M, Grandjean P, Auffhammer M, Sunding D. 2020. Reducing exposure to high levels of perfluorinated compounds in drinking water improves reproductive outcomes: evidence from an intervention in Minnesota. Environ Health 19(1):42, PMID: 32321520, https://doi.org/10.1186/s12940-020-00591-0.

Weinberg CR, Shi M, Basso O, DeRoo LA, Harmon Q, Wilcox AJ, et al. 2017. Season of conception, smoking, and preeclampsia in Norway. Environ Health Perspect 125(6):067022, PMID: 28669933, https://doi.org/10.1289/EHP963.

Wikström S, Lindh CH, Shu H, Bornehag C-G. 2019. Early pregnancy serum levels of perfluoroalkyl substances and risk of preeclampsia in Swedish women. Sci Rep 9(1):9179, PMID: 31235847, https://doi.org/10.1038/s41598-019-45483-7.

You S-H, Cheng P-J, Chung T-T, Kuo C-F, Wu H-M, Chu P-H. 2018. Population-based trends and risk factors of early- and late-onset preeclampsia in Taiwan 2001–2014. BMC Pregnancy Childbirth 18(1):199, PMID: 29855344, https://doi.org/10.1186/s12884-018-1845-7.

Zhang Y, Beesoon S, Zhu L, Martin JW. 2013. Biomonitoring of perfluoroalkyl acids in human urine and estimates of biological half-life. Environ Sci Technol 47(18):10619–10627, PMID: 23980546, https://doi.org/10.1021/es401905e.

Zhou Y, Damsky CH, Fisher SJ. 1997. Preeclampsia is associated with failure of human cytotrophoblasts to mimic a vascular adhesion phenotype. One cause of defective endovascular invasion in this syndrome? J Clin Invest 99(9):2152–2164, PMID: 9151787, https://doi.org/10.1172/JCI119388.